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Agricultural Products Research, Development and Testing

October 9, 1970

Mr. Morris Holman, P. E.
Chemical Leaman Trucking Lines
506 Lancaster Avenue
Dowington, Pa. 19335

Re: Analysis and Bioassay, Lagoon Waste
William Dick Property

Dear Mr. Holman:

We have now completed our bioassay experiments and limited chemical analyses on the sample of raw waste from lagoon #1 of the above source submitted personally by you on September 16, 1970 as well as the supernatant from a chemical flocculation of the same sample performed according to the procedure to be described below:

The raw waste was milky white in appearance. More complete chemical characterization was being done elsewhere, so that our analytical data are very limited. A four liter quantity of the sample was flocculated using a standard jar test apparatus by adding 1,000 ppm of alum and 200 ppm of lime in that sequence. Flocculation and settling was reasonably rapid, so that after about one hour a clear supernatant could readily be decanted for a few chemical assays and for bioassay experiments. The raw sample was assigned our laboratory #WW-289, and the supernatant from flocculation #WW-296. They will be identified by these numbers from this point.

Table I
Chemical Analysis*

	<u>WW-289</u>	<u>WW-296</u>
pH	7.1	5.5
Alkalinity	510	274
Acidity	54	35
Total Solids	3,866	1,734
Total Volatile Solids	3,256	735
Total Fixed Solids	610	999

*All values except pH, expressed as mg/l (ppm)

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Microbiology:

An extended range total plate count was made by our Microbiologist and found to be in excess of 30,000,000 viable microorganisms/ml, estimated 70,000,000. This high value precludes the presence of generally toxic conditions in the lagoon, otherwise even bacteria and similar microorganisms would not live.

Fish Bioassay:

Four adult guppies (2 male, 2 female) were placed in each of 10 small fish bowls containing 1500 ml quantities of our own raw well water, gentle aeration and light feeding every other day were provided for a three day period of acclimatization and for 14 days thereafter. After the acclimatization period appropriate quantities of the tap water were withdrawn and replaced with amounts of WW-289 or WW-296 to provide the concentrations desired. In Table II below are presented (1) an outline of the experimental parameters and (2) mortality data during the 14 day experimental period.

Table II
Guppie Bioassay Experiment

<u>Bowl#</u>	<u>Contents</u>	<u>Mortality</u>
1	Control	0
2	10,000 ppm, WW-289	3 (1 each at 3,4,& 8 days)
3	1,000 " "	1 (6 days)
4	100 " "	0
5	10 " "	1 (5 days)
6	Control	0
7	10,000 ppm, WW-296	1 (8 days)
8	1,000 " "	0
9	100 " "	0
10	10 " "	0

Bowl #2 after sample addition was extremely cloudy and foam developed due to the aeration. Its counterpart containing the high level of the supernatant was clear and, as indicated, only one death was noted.

Mouse Bioassay:

Into each of seven laboratory small animal cages were placed two white mice (1 male and 1 female). They were fed a diet of "Prime" dog food cubes and provided with laboratory tap water free choice for a 3 day orientation period. At that time the water supplies were changed to include the desired amounts of one or the other waste water. Table III offers information concerning experimental design and mortality data. Like the guppie test the experiment continued for 14 days.

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Table III
Mouse Bioassay Experiment

<u>Cage #</u>	<u>Water Composition</u>	<u>Mortality</u>
1	Control (tap water)	1 (12 days)
2	10,000 ppm - WW-289	1 (2 days)
3	1,000 " "	1 (8 days)
4	100 " "	0
5	10,000 " "	0
6	1,000 " "	0
7	100 " "	0

Possibly some explanation should be offered for a death having occurred in the control pen. Extremely warm weather was experienced during the first 6 days of the experiment, so that cages were kept in a room with windows wide open. A very sudden overnight change in weather (to a damp - cool) occurred so that during days 7-10 a general morbidity was noted in several of the mice. It is felt that the control death (and possibly the #3) occurred as a result of this condition. The death in cage 2 was, however, almost certainly a "toxic" death.

The data as offered do, I believe, speak for themselves in a general way. Obviously, chemical flocculation is effective in improving appearance and in providing a supernatant whose effect upon living organisms is minimal. In accordance with your request, treatability studies are now underway to determine whether a still more effective set of flocculation conditions can be developed.

Sincerely yours,

Earl H. Hess
Earl H. Hess, Ph.D.
President

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